

**APPLICATION**  
**FOR**  
**UNITED STATES LETTERS PATENT**

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**FOR**  
  
**SUSTAINED RELEASE PHARMACEUTICAL FORMULATION**  
**FOR INHALATION**

# **SUSTAINED RELEASE PHARMACEUTICAL FORMULATION FOR INHALATION**

## **Cross-Reference To Related Applications**

5           Priority is claimed to U.S. Provisional Application No. 60/414,951, filed September 30, 2002. The application is incorporated herein by reference.

## **Background of the Invention**

10           This invention is generally in the field of pharmaceutical formulations for delivery to the lungs by inhalation, and more particularly to microparticulate formulations for sustained release of pharmaceutical agents to the lungs.

          Delivery of pharmaceutical agents to the lungs and through the lungs to the body represents a large medical opportunity. Delivery of pharmaceutical agents to the lungs to treat respiratory ailments represents a large and growing medical need. Current pulmonary delivery systems are not ideal, often delivering inaccurate doses, requiring frequent dosing and losing significant amounts of pharmaceutical agent in the delivery process. For example, most asthma pharmaceutical agents delivered by inhalation are immediate release formulations that must be inhaled multiple times per day, which discourages patient compliance. In addition, frequent inhalation dosing of immediate release formulations leads to pharmaceutical agent levels that peak and trough, causing undesirable toxicity or inadequate efficacy.

20           Effective and efficient pulmonary pharmaceutical agent delivery presents significant technological challenges. To deliver pharmaceutical agents via inhalation, compounds must be precisely formulated to ensure that they are deposited to the appropriate part of the lung and to deliver the correct amount of pharmaceutical agent over the appropriate amount of time. This requires control of key factors such as geometric particle size and density and compatibility with select delivery devices.

25           Conventional efforts towards sustained release particles for inhalation have focused on the use of complexing agents, such as complexing a polycationic agent with a therapeutic agent. See, for example, U.S. Patent Application Publication No. 2003/0068277 A1 to Vanbever, et al. This approach, however, requires the therapeutic

agent to be able to form a complex with the polycationic agent, which limits the therapeutic agents to anionic compounds. This approach also requires the polycation complexing agent to be non-toxic to the lungs. This approach also has limited ability to control the release rate of the compound from the complex, as the release rate is essentially dependent upon the binding strength of the compound to the polycation.

Others have focused on designing formulations to target delivery to the deep lung, in order to avoid the mucociliary clearance mechanism and have the particle persist in the lungs for a longer duration. See, for example, U.S. Patent No. 6,060,069 to Hill et al. However, this approach cannot be used for delivery of pharmaceutical agents with therapeutic targets in the central and upper airways. In addition, this approach has limited ability to control the delivery rate, as it relies on the inherent dissolution rate of the pharmaceutical agent particles which will be governed primarily by the particle diameter and pharmaceutical agent solubility.

Others have focused on modulating release of a pharmaceutical agent delivered to the lung by varying matrix transition temperatures via the selective addition of a carboxylate moiety, a phospholipid and a multivalent salt or ionic components. See, for example, PCT WO 01/13891 to Basu et al. For slowing the release rate, materials with higher matrix transition temperatures are used. This approach is limited to pharmaceutical agents for which the highest matrix transition temperature materials provide sufficiently slow release.

It would be desirable to provide a sustained release, microparticle formulation of pharmaceutical agents, for local delivery to the lungs or systemic delivery via the lungs. It also would be desirable to provide a microparticle formulation of pharmaceutical agent enabling less frequent dosing, for example for efficacious once-daily dosing of a pharmaceutical agent useful in the treatment of asthma.

### **Summary of the Invention**

Pharmaceutical formulations and methods are provided for the sustained delivery of a pharmaceutical agent to the lungs of a patient by inhalation.

In one aspect, a sustained release pharmaceutical formulation is provided which comprises porous microparticles which comprise a pharmaceutical agent and a matrix

material, wherein upon inhalation of the formulation into the lungs a therapeutically or prophylactically effective amount of the pharmaceutical agent is released from the microparticles in the lungs for at least 2 hours (e.g., at least 4, 6, 8, 16, or 20 hours). In preferred embodiments, a majority of the pharmaceutical agent is released from the microparticles by 24 hours following inhalation. In one embodiment, a majority of the pharmaceutical agent is released no earlier than about 2 hours and no later than about 24 hours following inhalation (e.g., no earlier than about 6 hours and no later than about 18 hours, or no earlier than about 4 hours and no later than about 12 hours, etc.).

In one embodiment, the porous microparticles have a volume average diameter between about 1  $\mu\text{m}$  and 5  $\mu\text{m}$ . In another embodiment, the porous microparticles have a volume median diameter between about 1  $\mu\text{m}$  and 5  $\mu\text{m}$ . In one embodiment, the porous microparticles have an average porosity between about 15 and 90% by volume.

A variety of pharmaceutical agents can be employed in the pharmaceutical formulations. For example, the pharmaceutical agent can be a bronchodilator, a steroid, an antibiotic, an antiasthmatic, an antineoplastic, a peptide, or a protein. In one embodiment, the pharmaceutical agent comprises a corticosteroid, such as budesonide, fluticasone propionate, beclomethasone dipropionate, mometasone, flunisolide, and triamcinolone acetonide. In one embodiment, the sustained release formulation further comprises one or more other pharmaceutical agents.

In various embodiments, the matrix material is a biocompatible synthetic polymer, a lipid, a salt, a hydrophobic small molecule, or a combination thereof. Representative polymers include poly(hydroxy acids) such as poly(lactic acid), poly(glycolic acid), and poly(lactic acid-co-glycolic acid), poly(lactide), poly(glycolide), poly(lactide-co-glycolide), polyanhydrides, polyorthoesters, polyamides, polyalkylenes such as polyethylene and polypropylene, polyalkylene glycols such as poly(ethylene glycol), polyalkylene oxides such as poly(ethylene oxide), polyvinyl alcohols, polyvinyl ethers, polyvinylpyrrolidone, poly(butyric acid), poly(valeric acid), and poly(lactide-co-caprolactone), copolymers, derivatives, and blends thereof. In one embodiment, the polymer is a poly(lactide-co-glycolide) copolymerized with polyethylene glycol.

In one embodiment, the porous microparticles further comprise one or more

surfactants, such as a phospholipid.

In one embodiment, one or more pharmaceutically acceptable bulking agents are blended with the porous microparticles to form a dry powder blend formulation. The bulking agent can, for example, comprise particles which have a volume average size  
5 between 10 and 500  $\mu\text{m}$ . Examples of bulking agents include lactose, mannitol, sorbitol, trehalose, xylitol, and combinations thereof

In one embodiment, the formulations comprise one or more pharmaceutically acceptable suspending agents that are liquid within a metered dose inhaler to form a metered dose inhaler formulation.

10 In one embodiment, the sustained release formulation further comprises additional microparticles blended with the porous microparticles. For example, the additional microparticles can comprise one or more other pharmaceutical agents.

In one embodiment, at least 50% by weight of the microparticles delivered to the lung is delivered to the combined central and upper lung upon inhalation by the patient.

15 In one particular embodiment, a dry powder sustained release pharmaceutical formulation is provided which comprises porous microparticles having a volume average diameter between about 1  $\mu\text{m}$  and 5  $\mu\text{m}$ , the porous microparticles being formed at least of a pharmaceutical agent, a matrix material, and a surfactant, and a pharmaceutically acceptable bulking agent blended with the porous microparticles, wherein upon inhalation  
20 of the formulation into the lungs a majority of the pharmaceutical agent is released no earlier than about 2 hours and no later than about 24 hours following inhalation. In one embodiment, the patient orally inhales the sustained release formulation using a dry powder inhalation device.

In another aspect, a method of delivering a pharmaceutical agent to the lungs of a  
25 patient is provided. In one embodiment, the method comprises having the patient inhale a sustained release pharmaceutical formulation which comprises porous microparticles which comprise a pharmaceutical agent and a matrix material, wherein upon inhalation of the formulation into the lungs a therapeutically or prophylactically effective amount of the pharmaceutical agent is released from the microparticles in the lungs for at least 2 hours  
30 (e.g., at least 4, 8, or 16 hours). In preferred embodiments, a majority of the

pharmaceutical agent is released from the microparticles by 24 hours following inhalation (e.g., no earlier than about 10 hours and no later than about 24 hours, or no earlier than about 6 hours and no later than about 18 hours, etc.).

5 In one embodiment, the patient is in need of treatment for a respiratory disease or disorder, such as asthma. In various embodiments of the method, the pharmaceutical agent, such as a corticosteroid, is released over a duration that extends up to at least about 2 hours, and preferably completes release by about 24 hours (e.g., the majority of the pharmaceutical agent is released between about 4 and about 24 hours, between about 8 and about 24 hours, between about 10 and about 24 hours, between about 6 and about 18 hours, 10 or between about 4 and about 12 hours).

In one embodiment, the method and formulation provide local or plasma concentrations at approximately constant values which do not fluctuate by more than a factor of four over the period of sustained release. In another embodiment, a sustained release pharmaceutical formulation for delivery to the lungs of a patient by inhalation 15 comprising: porous microparticles which comprise a pharmaceutical agent and a matrix material, wherein upon inhalation of the formulation into the lungs there is an increase in  $MAT_{inh}$  of at least 25% compared to the  $MAT_{inh}$  obtained when the pharmaceutical agent is administered by inhalation of microparticles not in the form of porous microparticles which comprise the pharmaceutical agent and the matrix material.

20 In another aspect, a method for making a dry powder formulation for inhalation and sustained release of a pharmaceutical agent is provided. In one embodiment, the method comprises dissolving a matrix material in a volatile solvent to form a solution; adding a pharmaceutical agent to the solution to form an emulsion, suspension, or second solution; and removing the volatile solvent from the emulsion, suspension, or second solution to 25 yield porous microparticles which comprise the pharmaceutical agent and the matrix material, wherein upon inhalation of the formulation into the lungs a therapeutically or prophylactically effective amount of the pharmaceutical agent is released from the microparticles in the lungs for at least 2 hours. In one embodiment, the matrix material comprises a biocompatible synthetic polymer, and the volatile solvent comprises an

organic solvent. In another embodiment, the method further comprises combining one or more surfactants, such as a phospholipid, with the solution.

In another embodiment, the method includes dissolving a matrix material, and optionally a surfactant, in a volatile solvent to form a solution, combining a pharmaceutical agent with the matrix material solution; combining at least one pore forming agent with the pharmaceutical agent in the matrix solution to form an emulsion, suspension, or second solution; and removing the volatile solvent and the pore forming agent from the emulsion, suspension, or second solution to yield porous microparticles which comprise the pharmaceutical agent and the matrix material, wherein upon inhalation of the formulation into the lungs a therapeutically or prophylactically effective amount of the pharmaceutical agent is released from the microparticles in the lungs for at least 2 hours. In one embodiment, the pore forming agent (e.g., a volatile salt) is in the form of an aqueous solution when combined with the pharmaceutical agent in the matrix solution. In one embodiment, the step of removing the volatile solvent and pore forming agent from the emulsion, suspension, or second solution is conducted using a process selected from spray drying, evaporation, fluid bed drying, lyophilization, vacuum drying, or a combination thereof. In another embodiment, the method further comprises blending the porous microparticles with a pharmaceutically acceptable bulking agent.

#### **Brief Description of the Drawings**

FIG. 1 is a graph of percent *in vitro* release of budesonide after 5.5 hours versus percent porosity of the microparticles.

FIG. 2 is a graph of percent *in vitro* release of fluticasone propionate after 5.5 hours versus percent porosity of the microparticles.

FIG. 3 is a graph of percent *in vitro* release of fluticasone propionate after 24 hours versus percent porosity of the microparticles.

FIG. 4 is a graph showing plasma profiles of budesonide (adjusted for actual inhaled dose) over time following dosing, comparing a commercially available immediate release formulation (Pulmicort) versus one embodiment of a sustained release formulation comprising porous microparticles described herein.

### **Detailed Description of the Invention**

A sustained release delivery system for pharmaceutical agents delivered locally to the lung or for pharmaceutical agents delivered systemically through the lungs, has been developed. The delivery system is a formulation comprising porous microparticles, where porosity, particle geometric diameter and composition are selected and used to control the rate of release of pharmaceutical agent from the microparticles following inhalation into the lungs. In particular, it has been discovered that the composition of the microparticles (e.g., the matrix material, surfactant) can be selected to provide delayed release (and avoid the burst effect associated with immediate release formulations), and the porosity of the microparticles can be selected to provide the majority of the pharmaceutical agent release before the microparticles are removed by the pulmonary clearance mechanisms. Although the composition of the microparticles can be selected to slow the release of the pharmaceutical agent, selection of the composition alone may not ensure that a sufficient amount of pharmaceutical agent is released before the microparticles are removed by the pulmonary clearance mechanisms. For a given composition of the microparticles, the porosity can be selected to ensure that a therapeutically or prophylactically effective amount of the pharmaceutical agent continues to be released after 2 hours, preferably such that a majority (e.g., more than 50%, more than 75%, more than 90% by weight of the pharmaceutical agent) of the pharmaceutical agent is released from the microparticles by 24 hours following inhalation.

Advantageously, the porous microparticles can provide sustained local delivery of pharmaceutical agent and/or sustained plasma levels without the need to complex the pharmaceutical agent molecule with another molecule. In addition, the sustained delivery formulations advantageously can moderate the pharmaceutical agent peaks and troughs associated with immediate release pharmaceutical agents, which can cause added toxicity or reduced efficacy.

Advantageously, the sustained release formulations can deliver a majority of the inhaled microparticles to the appropriate region of the lung for the desired therapeutic or prophylactic use. That is, preferably, at least 50% by weight of the microparticles delivered to the lung is delivered, upon inhalation by the patient, to the appropriate region



of the lung (for example, the combined central and upper lung) for the desired therapeutic or prophylactic use.

Advantageously, the method and formulation can provide local or plasma concentrations at approximately constant values. For example, they may not fluctuate by more than a factor of four over the period of sustained release.

As used herein, the terms “comprise,” “comprising,” “include,” and “including” are intended to be open, non-limiting terms, unless the contrary is expressly indicated.

### **The Sustained Release Formulations**

The sustained release pharmaceutical formulations for pulmonary administration include porous microparticles that comprise a pharmaceutical agent and a matrix material. The microparticle's composition, geometric diameter, and porosity provide that upon inhalation of the formulation into the lungs a therapeutically or prophylactically effective amount of the pharmaceutical agent is released in a sustained manner from the microparticles in the lungs over a duration that extends up to at least about 2 hours, and preferably completes release by about 24 hours.

As a measure of sustained release, the mean absorption time following inhalation ( $MAT_{inh}$ ) for the drug can be used. The  $MAT_{inh}$  is the average time it takes for a drug molecule to be absorbed into the bloodstream from the lungs following inhalation and can be calculated from the pharmaceutical agent plasma profile following inhalation as follows:

$$MAT_{inh} = (AUMC_{inh\infty}/AUC_{inh\infty}) - MRT_{iv} \quad (EQ.1)$$

where  $AUMC_{inh\infty}$  is area under the first moment curve (product of time and plasma concentration) from time zero to infinity following inhalation,  $AUC_{inh\infty}$  is the area under the plasma concentration curve from time zero to infinity following inhalation, and  $MRT_{iv}$  is the mean residence time for the pharmaceutical agent of interest following intravenous administration. The  $MRT_{iv}$  can be determined as follows:

$$MRT_{iv} = (AUMC_{iv\infty}/AUC_{iv\infty}) \quad (EQ.2)$$

where  $AUMC_{iv\infty}$  is area under the first moment curve (product of time and plasma concentration) from time zero to infinity following intravenous administration, and  $AUC_{iv\infty}$  is the area under the plasma concentration curve from time zero to infinity following

intravenous administration.

For example, the porous microparticles can provide a pharmaceutical agent mean absorption time following inhalation greater than the pharmaceutical agent mean absorption time following inhalation when not delivered in microparticle form. The desired  
5 MAT<sub>inh</sub> will depend on the drug molecule to be administered, and it is helpful to consider the increase in MAT<sub>inh</sub> obtained using the present microparticle formulations compared to the drug molecule when not delivered as microparticles. In preferred embodiments, a drug administered in microparticles of the present compositions and methods will provide an increase in MAT<sub>inh</sub> of at least between about 25 and 50% as compared to the drug  
10 administered not in the present microparticles.

The sustained release formulations are achieved by controlling microparticle composition, microparticle geometric size, and microparticle porosity. Porosity ( $\epsilon$ ) is the ratio of the volume of voids contained in the microparticles ( $V_v$ ) to the total volume of the microparticles ( $V_t$ ):

$$15 \quad \epsilon = V_v/V_t \quad (\text{EQ.3})$$

This relationship can be expressed in terms of the envelope density ( $\rho_e$ ) of the microparticles and the absolute density ( $\rho_a$ ) of the microparticles:

$$\epsilon = 1 - \rho_e / \rho_a \quad (\text{EQ.4})$$

The absolute density is a measurement of the density of the solid material present in the  
20 microparticles, and is equal to the mass of the microparticles (which is assumed to equal the mass of solid material, as the mass of voids is assumed to be negligible) divided by the volume of the solid material (i.e., excludes the volume of voids contained in the microparticles and the volume between the microparticles). Absolute density can be measured using techniques such as helium pycnometry. The envelope density is equal to  
25 the mass of the microparticles divided by the volume occupied by the microparticles (i.e., equals the sum of the volume of the solid material and the volume of voids contained in the microparticles and excludes the volume between the microparticles). Envelope density can be measured using techniques such as mercury porosimetry or using a GeoPyc<sup>TM</sup> instrument (Micromeritics, Norcross, Georgia). However, such methods are limited to  
30 geometric particle sizes larger than desirable for pulmonary applications. The envelope

density can be estimated from the tap density of the microparticles. The tap density is a measurement of the packing density and is equal to the mass of microparticles divided by the sum of the volume of solid material in the microparticles, the volume of voids within the microparticles, and the volume between the packed microparticles of the material. Tap density ( $\rho_t$ ) can be measured using a GeoPyc<sup>TM</sup> instrument or techniques such as those described in the British Pharmacopoeia and ASTM standard test methods for tap density. It is known in the art that the envelope density can be estimated from the tap density for essentially spherical microparticles by accounting for the volume between the microparticles:

$$\rho_e = \rho_t / 0.794 \quad (\text{EQ.5})$$

The porosity can be expressed as follows:

$$\varepsilon = 1 - \rho_t / (0.794 * \rho_a) \quad (\text{EQ.6})$$

For a given microparticle composition (pharmaceutical agent and matrix material) and structure (microparticle porosity and thus density) an iterative process can be used to define where the microparticles go in the lung and the duration over which the microparticles release the pharmaceutical agent: (1) the matrix material, the pharmaceutical agent content, and the microparticle geometric size are selected to determine the time and amount of initial pharmaceutical agent release; (2) the porosity of the microparticles is selected to adjust the amount of initial pharmaceutical agent release, and to ensure that significant release of the pharmaceutical agent occurs beyond the initial release and that the majority of the pharmaceutical agent release occurs within 24 hours; and then (3) the geometric particle size and the porosity are adjusted to achieve a certain aerodynamic diameter which enables the particles to be deposited by inhalation to the region of interest in the lung. As used herein, the term “initial release” refers to the amount of pharmaceutical agent released shortly after the microparticles become wetted. The initial release upon wetting of the microparticles results from pharmaceutical agent which is not fully encapsulated and/or pharmaceutical agent which is located close to the exterior surface of the microparticle. The amount of pharmaceutical agent released in the first 10 minutes is used as a measure of the initial release.

As used herein, the terms “diameter” or “d” in reference to particles refers to the

number average particle size, unless otherwise specified. An example of an equation that can be used to describe the number average particle size is shown below:

$$d = \frac{\sum_{i=1}^p n_i d_i}{\sum_{i=1}^p n_i} \quad (\text{EQ.7})$$

where n = number of particles of a given diameter (d).

5 As used herein, the terms "geometric size," "geometric diameter," "volume average size," "volume average diameter" or "d<sub>g</sub>" refers to the volume weighted diameter average. An example of equations that can be used to describe the volume average diameter is shown below:

$$d_g = \left[ \frac{\sum_{i=1}^p n_i d_i^3}{\sum_{i=1}^p n_i} \right]^{1/3} \quad (\text{EQ.8})$$

10 where n = number of particles of a given diameter (d).

As used herein, the term "volume median" refers to the median diameter value of the volume-weighted distribution. The median is the diameter for which 50% of the total are smaller and 50% are larger, and corresponds to a cumulative fraction of 50%.

15 Geometric particle size analysis can be performed on a Coulter counter, by light scattering, by light microscopy, scanning electron microscopy, or transmittance electron microscopy, as known in the art.

As used herein, the term "aerodynamic diameter" refers to the equivalent diameter of a sphere with density of 1 g/mL were it to fall under gravity with the same velocity as the particle analyzed. The aerodynamic diameter (d<sub>a</sub>) of a microparticle is related to the geometric diameter (d<sub>g</sub>) and the envelope density (ρ<sub>e</sub>) by the following:

$$d_a = d_g \sqrt{\rho_e} \quad (\text{EQ.9})$$

Porosity affects envelope density (EQ. 4) which in turn affects aerodynamic diameter.

Thus porosity can be used to affect both where the microparticles go in the lung and the rate at which the microparticles release the pharmaceutical agent in the lung. Gravitational

25 settling (sedimentation), inertial impaction, Brownian diffusion, interception and

electrostatic precipitation affect particle deposition in the lungs. Gravitational settling and inertial impaction are dependent on  $d_a$  and are the most important factors for deposition of particles with aerodynamic diameters between 1  $\mu\text{m}$  and 10  $\mu\text{m}$ . Particles with  $d_a > 10 \mu\text{m}$  will not penetrate the tracheobronchial tree, particles with  $d_a$  in the 3-10  $\mu\text{m}$  range have  
5 predominantly tracheobronchial deposition, particles with  $d_a$  in the 1-3  $\mu\text{m}$  range are deposited in the alveolar region (deep lung), and particles with  $d_a < 1 \mu\text{m}$  are mostly exhaled. Respiratory patterns during inhalation can shift these aerodynamic particle size ranges slightly. For example, with rapid inhalation, the tracheobronchial region shifts to between 3  $\mu\text{m}$  and 6  $\mu\text{m}$ . It is a generally held belief that the ideal scenario for delivery to  
10 the lung is to have  $d_a < 5 \mu\text{m}$ . See, e.g., Edwards et al., *J. Appl. Physiol.* 85(2):379-85 (1998); Suarez & Hickey, *Respir. Care*, 45(6):652-66 (2000).

Aerodynamic particle size analysis can be performed via cascade impaction, liquid impinger analysis, or time-of-flight methods, as known in the art.

#### The Porous Microparticles

15 The porous microparticles comprise a matrix material and a pharmaceutical agent. As used herein, the term “matrix” refers to a structure including one or more materials in which the pharmaceutical agent is dispersed, entrapped, or encapsulated. The matrix is in the form of porous microparticles. Optionally, the porous microparticles further include one or more surfactants.

20 As used herein, the term “microparticle” includes microspheres and microcapsules, as well as microparticles, unless otherwise specified. Microparticles may or may not be spherical in shape. Microcapsules are defined as microparticles having an outer shell surrounding a core containing another material, for example, the pharmaceutical agent. Microspheres comprising pharmaceutical agent and matrix can be porous having a  
25 honeycombed structure or a single internal void. Either type of microparticle may also have pores on the surface of the microparticle.

In one embodiment, the microparticles have a volume average diameter between 0.1 and 5  $\mu\text{m}$  (e.g., between 1 and 5  $\mu\text{m}$ , between 2 and 5  $\mu\text{m}$ , etc.). In another embodiment, the microparticles have a volume average diameter of up to 10  $\mu\text{m}$ , for  
30 targeting delivery to the large bronchi. Particle size (geometric diameter and aerodynamic

diameter) is selected to provide an easily dispersed powder that upon aerosolization and inhalation readily deposits at a targeted site in the respiratory tract (e.g., upper airway, deep lung, etc.), preferably while avoiding or minimizing excessive deposition of the particles in the oropharynx or nasal regions. In one preferred embodiment, the porous microparticles have a volume average diameter of between 2 and 5  $\mu\text{m}$ . The volume average diameter is also selected to avoid and minimize effects of one of the lung's natural clearance mechanisms (e.g. phagocytosis by macrophages). Generally, larger particles are phagocytosed at a slower rate.

In one embodiment, the microparticles have an average porosity between about 15 and 90%. The porosity of the microparticles is selected so that the majority of the pharmaceutical agent is released before the particle is removed from the lung by biological clearance mechanisms such as mucociliary clearance. In specific embodiments, the average porosity can be between about 25 and about 75%, between about 35 and about 65%, or between about 40 and about 60%.

#### Matrix Material

The matrix material is a material that functions to slow down release of the pharmaceutical agent from the microparticle. It can be formed of non-biodegradable or biodegradable materials, although biodegradable materials are preferred, particularly for inhalation administration.

The matrix material can be crystalline, semi-crystalline, or amorphous. The matrix material may be a polymer, a lipid, a salt, a hydrophobic small molecule, or a combination thereof.

The pharmaceutical agent can be present in the porous microparticle in an amount that is greater than or less than the amount of matrix material that is present in the porous microparticle, depending upon the particular formulation needs.

The matrix material comprises at least 5%w/w of the microparticle. The content of matrix material in the microparticles can be between 5 and about 95 wt%. In typical embodiments, the matrix material is present in an amount between about 50 and 90 wt%.

Representative synthetic polymers include poly(hydroxy acids) such as poly(lactic acid), poly(glycolic acid), and poly(lactic acid-co-glycolic acid), poly(lactide),

poly(glycolide), poly(lactide-co-glycolide), polyanhydrides, polyorthoesters, polyamides, polyalkylenes such as polyethylene and polypropylene, polyalkylene glycols such as poly(ethylene glycol), polyalkylene oxides such as poly(ethylene oxide), polyvinyl alcohols, polyvinyl ethers, polyvinylpyrrolidone, poly(butyric acid), poly(valeric acid), and poly(lactide-co-caprolactone), copolymers, derivatives, and blends thereof. As used herein, "derivatives" include polymers having substitutions, additions of chemical groups, for example, alkyl, alkylene, hydroxylations, oxidations, and other modifications routinely made by those skilled in the art.

Examples of preferred biodegradable polymers include polymers of hydroxy acids such as lactic acid and glycolic acid (including poly(lactide-co-glycolide)), and copolymers with PEG, polyanhydrides, poly(ortho)esters, poly(butyric acid), poly(valeric acid), poly(lactide-co-caprolactone), blends and copolymers thereof.

Examples of preferred natural polymers include proteins such as albumin, fibrinogen, gelatin, and prolamines, for example, zein, and polysaccharides such as alginate, cellulose and polyhydroxyalkanoates, for example, polyhydroxybutyrate.

Representative lipids include the following classes of molecules: fatty acids and derivatives, mono-, di- and triglycerides, phospholipids, sphingolipids, cholesterol and steroid derivatives, terpenes, and vitamins. Fatty acids and derivatives thereof may include saturated and unsaturated fatty acids, odd and even number fatty acids, cis and trans isomers, and fatty acid derivatives including alcohols, esters, anhydrides, hydroxy fatty acids and prostaglandins. Saturated and unsaturated fatty acids that may be used include molecules that have between 12 carbon atoms and 22 carbon atoms in either linear or branched form. Examples of saturated fatty acids that may be used include lauric, myristic, palmitic, and stearic acids. Examples of unsaturated fatty acids that may be used include lauric, physeteric, myristoleic, palmitoleic, petroselinic, and oleic acids. Examples of branched fatty acids that may be used include isolauric, isomyristic, isopalmitic, and isostearic acids and isoprenoids. Fatty acid derivatives include 12-(((7'-diethylaminocoumarin-3-yl)carbonyl)methylamino)-octadecanoic acid; N-[12-(((7'-diethylaminocoumarin-3-yl) carbonyl)methyl-amino) octadecanoyl]-2-aminopalmitic acid, N succinyl-dioleoylphosphatidylethanol amine and palmitoyl-homocysteine; and/or

combinations thereof. Mono, di- and triglycerides or derivatives thereof that may be used include molecules that have fatty acids or mixtures of fatty acids between 6 and 24 carbon atoms, digalactosyldiglyceride, 1,2-dioleoyl-sn-glycerol; 1,2-dipalmitoyl-sn-3 succinylglycerol; and 1,3-dipalmitoyl-2-succinylglycerol.

5 In one preferred embodiment, the matrix material comprises a phospholipid or combinations of phospholipids. Phospholipids that may be used include phosphatidic acids, phosphatidyl cholines with both saturated and unsaturated lipids, phosphatidyl ethanolamines, phosphatidylglycerols, phosphatidylserines, phosphatidylinositols, lysophosphatidyl derivatives, cardiolipin, and  $\beta$ -acyl- $\gamma$ -alkyl phospholipids. Examples of  
10 phosphatidylcholines include such as dioleoylphosphatidylcholine, dimyristoylphosphatidylcholine (DMPC), dipentadecanoylphosphatidylcholine dilauroylphosphatidylcholine, dipalmitoylphosphatidylcholine (DPPC), distearoylphosphatidylcholine (DSPC), diarachidoylphosphatidylcholine (DAPC), dibehenoylphosphatidylcholine (DBPC), ditricosanoylphosphatidylcholine (DTPC),  
15 dilignoceroylphatidylcholine (DLPC); and phosphatidylethanolamines such as dioleoylphosphatidylethanolamine or 1-hexadecyl-2-palmitoylglycerophosphoethanolamine. Synthetic phospholipids with asymmetric acyl chains (e.g., with one acyl chain of 6 carbons and another acyl chain of 12 carbons) may also be used. Examples of phosphatidylethanolamines include  
20 dicaprylphosphatidylethanolamine, dioctanoylphosphatidylethanolamine, dilauroylphosphatidylethanolamine, dimyristoylphosphatidylethanolamine (DMPE), dipalmitoylphosphatidylethanolamine (DPPE), dipalmitoleoylphosphatidylethanolamine, distearoylphosphatidylethanolamine (DSPE), dioleoylphosphatidylethanolamine, and dilineoylphosphatidylethanolamine. Examples of phosphatidylglycerols include  
25 dicaprylphosphatidylglycerol, dioctanoylphosphatidylglycerol, dilauroylphosphatidylglycerol, dimyristoylphosphatidylglycerol (DMPG), dipalmitoylphosphatidylglycerol (DPPG), dipalmitoleoylphosphatidylglycerol, distearoylphosphatidylglycerol (DSPG), dioleoylphosphatidylglycerol, and dilineoylphosphatidylglycerol. Preferred phospholipids include DMPC, DPPC, DAPC,  
30 DSPC, DTPC, DBPC, DMPG, DPPG, DSPG, DMPE, DPPE, and DSPE.



Additional examples of phospholipids include modified phospholipids for example phospholipids having their head group modified, e.g., alkylated or polyethylene glycol (PEG)-modified, hydrogenated phospholipids, phospholipids with multifarious head groups (phosphatidylmethanol, phosphatidylethanol, phosphatidylpropanol, 5 phosphatidylbutanol, etc.), dibromo phosphatidylcholines, mono and diphytanoly phosphatides, mono and diacetylenic phosphatides, and PEG phosphatides.

Sphingolipids that may be used include ceramides, sphingomyelins, cerebroside, gangliosides, sulfatides and lysosulfatides. Examples of sphingolipids include the gangliosides GM1 and GM2.

10 Steroids which may be used include cholesterol, cholesterol sulfate, cholesterol hemisuccinate, 6-(5-cholesterol 3 $\beta$ -yloxy) hexyl-6-amino-6-deoxy-1-thio- $\alpha$ -D-galactopyranoside, 6-(5-cholesten-3  $\beta$ -yloxy)hexyl-6-amino-6-deoxyl-1-thio- $\alpha$ -D mannopyranoside and cholesteryl(4'-trimethyl 35 ammonio)butanoate.

Additional lipid compounds that may be used include tocopherol and derivatives, 15 and oils and derivatized oils such as stearlyamine.

Other suitable hydrophobic compounds include amino acids such as tryptophane, tyrosine, isoleucine, leucine, and valine, aromatic compounds such as an alkyl paraben, for example, methyl paraben, tyloxapol, and benzoic acid.

The matrix may comprise pharmaceutically acceptable small molecules such as 20 carbohydrates (including mono and disaccharides, sugar alcohols and derivatives of carbohydrates such as esters), and amino acids, their salts and their derivatives such as esters and amides.

A variety of cationic lipids such as DOTMA, N-[1-(2,3-dioleoyloxy)propyl-N,N,N-trimethylammonium chloride; DOTAP, 1,2-dioleoyloxy-3-(trimethylammonio) propane; 25 and DOTB, 1,2-dioleoyl-3-(4'-trimethyl-ammonio) butanoyl-sn glycerol may be used.

Inorganic materials can be included in the microparticles. Salts of metals (inorganic salts), such as calcium chloride or sodium chloride may be present in the particle or used in the production of the particles. Metal ions such calcium, magnesium, aluminum, zinc, sodium, potassium, lithium and iron may be used as the counterion for salts with organic 30 acids such as citric acid and/ or lipids including phospholipids. Examples of salts of

organic acids include sodium citrate, sodium ascorbate, magnesium gluconate, and sodium gluconate. A variety of metal ions may be used in such complexes, including lanthanides, transition metals, alkaline earth metals, and mixtures of metal ions. Salts of organic bases may be included such as tromethamine hydrochloride.

5 In one embodiment, the microparticles may include one or more carboxylic acid as the free acid or the salt form. The salt can be a divalent salt. The carboxylate moiety can be a hydrophilic carboxylic acid or salt thereof. Suitable carboxylic acids include hydroxydicarboxylic acids, hydroxytricarboxylic acids and the like. Citric acid and citrate are preferred. Suitable counterions for salts include sodium and alkaline earth metals such  
10 as calcium. Such salts can be formed during the preparation of the particles, from the combination of one type of salt such as calcium chloride and carboxylic acid as the free acid or an alternative salt form such as the sodium salt.

### Surfactants

In one embodiment, the porous microparticles further includes one or more  
15 surfactants. As used herein, a “surfactant” is a compound that is hydrophobic or amphiphilic (i.e., including both a hydrophilic and a hydrophobic component or region). Surfactants can be used to facilitate microparticle formation, to modify the surface properties of the microparticles and alter the way in which the microparticles are dispersed with a dry powder inhalation device or a metered dose inhaler, to alter the properties of the  
20 matrix material (e.g. to increase or decrease the hydrophobicity of the matrix), or to perform a combination of functions thereof. It is to be distinguished from similar or identical materials forming the “matrix material.” The content of surfactant in the porous microparticles generally is less than about 10% by weight of the microparticles.

In one embodiment, the surfactant comprises a lipid. Lipids that may be used  
25 include the following classes of lipids: fatty acids and derivatives, mono-, di- and triglycerides, phospholipids, sphingolipids, cholesterol and steroid derivatives, terpenes, prostaglandins and vitamins. Fatty acids and derivatives thereof may include saturated and unsaturated fatty acids, odd and even number fatty acids, cis and trans isomers, and fatty acid derivatives including alcohols, esters, anhydrides, hydroxy fatty acids, and salts of  
30 fatty acids. Saturated and unsaturated fatty acids that may be used include molecules that

have between 12 carbon atoms and 22 carbon atoms in either linear or branched form. Examples of saturated fatty acids that may be used include lauric, myristic, palmitic, and stearic acids. Examples of unsaturated fatty acids that may be used include lauric, physeteric, myristoleic, palmitoleic, petroselinic, and oleic acids. Examples of branched fatty acids that may be used include isolauric, isomyristic, isopalmitic, and isostearic acids and isoprenoids. Fatty acid derivatives include 12-(((7'-diethylaminocoumarin-3-yl)carbonyl)methylamino)-octadecanoic acid; N-[12-(((7'-diethylaminocoumarin-3-yl)carbonyl)methyl-amino) octadecanoyl]-2-aminopalmitic acid, N succinyl-dioleoylphosphatidylethanol amine and palmitoyl-homocysteine; and/or combinations thereof. Mono, di- and triglycerides or derivatives thereof that may be used include molecules that have fatty acids or mixtures of fatty acids between 6 and 24 carbon atoms, digalactosyldiglyceride, 1,2-dioleoyl-sn-glycerol; 1,2-dipalmitoyl-sn-3 succinylglycerol; and 1,3-dipalmitoyl-2-succinylglycerol.

In one preferred embodiment, the surfactant comprises a phospholipid.

Phospholipids that may be used include phosphatidic acids, phosphatidyl cholines with both saturated and unsaturated lipids, phosphatidyl ethanolamines, phosphatidylglycerols, phosphatidylserines, phosphatidylinositols, lysophosphatidyl derivatives, cardiolipin, and  $\beta$ -acyl- $\gamma$ -alkyl phospholipids. Examples of phosphatidylcholines include such as dioleoylphosphatidylcholine, dimyristoylphosphatidylcholine (DMPC), dipentadecanoylphosphatidylcholine dilauroylphosphatidylcholine, dipalmitoylphosphatidylcholine (DPPC), distearoylphosphatidylcholine (DSPC), diarachidoylphosphatidylcholine (DAPC), dibehenoylphosphatidylcholine (DBPC), ditricosanoylphosphatidylcholine (DTPC), dilignoceroylphatidylcholine (DLPC); and phosphatidylethanolamines such as dioleoylphosphatidylethanolamine or 1-hexadecyl-2--palmitoylglycerophosphoethanolamine. Synthetic phospholipids with asymmetric acyl chains (e.g., with one acyl chain of 6 carbons and another acyl chain of 12 carbons) may also be used. Examples of phosphatidylethanolamines include dicaprylphosphatidylethanolamine, dioctanoylphosphatidylethanolamine, dilauroylphosphatidylethanolamine, dimyristoylphosphatidylethanolamine (DMPE), dipalmitoylphosphatidylethanolamine (DPPE), dipalmitoleoylphosphatidylethanolamine,

distearoylphosphatidylethanolamine (DSPE), dioleoylphosphatidylethanolamine, and dilineoylphosphatidylethanolamine. Examples of phosphatidylglycerols include dicaprylphosphatidylglycerol, dioctanoylphosphatidylglycerol, dilauroylphosphatidylglycerol, dimyristoylphosphatidylglycerol (DMPG),  
5 dipalmitoylphosphatidylglycerol (DPPG), dipalmitoleoylphosphatidylglycerol, distearoylphosphatidylglycerol (DSPG), dioleoylphosphatidylglycerol, and dilineoylphosphatidylglycerol. Preferred phospholipids include DMPC, DPPC, DAPC, DSPC, DTPC, DBPC, DLPC, DMPG, DPPG, DSPG, DMPE, DPPE, and DSPE, and most preferably DPPC, DAPC and DSPC.

10 Sphingolipids that may be used include ceramides, sphingomyelins, cerebrosides, gangliosides, sulfatides and lysosulfatides. Examples of sphingolipids include the gangliosides GM1 and GM2.

Steroids which may be used include cholesterol, cholesterol sulfate, cholesterol hemisuccinate, 6-(5-cholesterol 3 $\beta$ -yloxy) hexyl-6-amino-6-deoxy-1-thio- $\alpha$ -D-  
15 galactopyranoside, 6-(5-cholesten-3  $\beta$ -yloxy)hexyl-6-amino-6-deoxyl-1-thio- $\alpha$ -D mannopyranoside and cholesteryl(4'-trimethyl 35 ammonio)butanoate.

Additional lipid compounds that may be used include tocopherol and derivatives, and oils and derivatized oils such as stearlyamine.

A variety of cationic lipids such as DOTMA, N-[1-(2,3-dioleoyloxy)propyl-N,N,N-  
20 trimethylammonium chloride; DOTAP, 1,2-dioleoyloxy-3-(trimethylammonio) propane; and DOTB, 1,2-dioleoyl-3-(4'-trimethyl-ammonio) butanoyl-sn glycerol may be used.

A variety of other surfactants may be used including ethoxylated sorbitan esters, sorbitan esters, fatty acid salts, sugar esters, pluronics, tetronics, ethylene oxides, butylene oxides, propylene oxides, anionic surfactants, cationic surfactants, mono and diacyl  
25 glycerols, mono and diacyl ethylene glycols, mono and diacyl sorbitols, mono and diacyl glycerol succinates, alkyl acyl phosphatides, fatty alcohols, fatty amines and their salts, fatty ethers, fatty esters, fatty amides, fatty carbonates, cholesterol esters, cholesterol amides and cholesterol ethers.

Examples of anionic or cationic surfactants include aluminum monostearate,  
30 ammonium lauryl sulfate, calcium stearate, dioctyl calcium sulfosuccinate, dioctyl

potassium sulfosuccinate, dioctyl sodium sulfosuccinate, emulsifying wax, magnesium lauryl sulfate, potassium oleate, sodium castor oil, sodium cetostearyl sulfate, sodium lauryl ether sulfate, sodium lauryl sulfate, sodium lauryl sulfoacetate, sodium oleate, sodium stearate, sodium stearyl fumarate, sodium tetradecyl sulfate, zinc oleate, zinc stearate, benzalconium chloride, cetrinide, cetrinide bromide, and cetylpyridinium chloride.

### Pharmaceutical Agent

A wide variety of pharmaceutical agents can be loaded within the porous microparticles of the sustained release formulations described herein. The “pharmaceutical agent” is a therapeutic, diagnostic, or prophylactic agent. It may be referred to herein generally as a “drug” or “active agent.” The pharmaceutical agent can be, for example, a protein, peptide, sugar, oligosaccharide, nucleic acid molecule, or other synthetic or natural agent. The pharmaceutical agent may be present in an amorphous state, a crystalline state, or a mixture thereof.

Representative examples of suitable pharmaceutical agents include the following categories and examples of pharmaceutical agents and alternative forms of these pharmaceutical agents such as alternative salt forms, free acid forms, free base forms, and hydrates:

analgesics/antipyretics (e.g., aspirin, acetaminophen, ibuprofen, naproxen sodium, buprenorphine, propoxyphene hydrochloride, propoxyphene napsylate, meperidine hydrochloride, hydromorphone hydrochloride, morphine, oxycodone, codeine, dihydrocodeine bitartrate, pentazocine, hydrocodone bitartrate, levorphanol, diflunisal, trolamine salicylate, nalbuphine hydrochloride, mefenamic acid, butorphanol, choline salicylate, butalbital, phenyltoloxamine citrate, diphenhydramine citrate, methotrimeprazine, cinnamedrine hydrochloride, fentanyl, and meprobamate);

antiasthmatics (e.g., xanthines such as theophylline, aminophylline, dyphylline, metaproterenol sulfate, and aminophylline; mast cell stabilizers such as cromolyn sodium and nedocromil sodium; anticholinergic agents such as ipratropium bromide; inhalant corticosteroids such as budesonide, beclomethasone dipropionate, flunisolide, triamcinolone acetonide, mometasone, and fluticasone propionate; leukotriene modifiers

- such as zafirlukast and zileuton; corticosteroids such as methyl prednisolone, prednisolone, prednisone, ketotifen, and traxanox);
- antibiotics (e.g., neomycin, streptomycin, chloramphenicol, cephalosporin, ampicillin, penicillin, tetracycline, and ciprofloxacin);
- 5 antidepressants (e.g., nefopam, oxypertine, doxepin, amoxapine, trazodone, amitriptyline, maprotiline, phenelzine, desipramine, nortriptyline, tranylcypromine, fluoxetine, imipramine, imipramine pamoate, isocarboxazid, trimipramine, and protriptyline);
- antidiabetics (e.g., biguanides and sulfonylurea derivatives);
- antifungal agents (e.g., griseofulvin, ketoconazole, itraconazole, amphotericin B, nystatin, voriconazole, and candicidin);
- 10 antihypertensive agents (e.g., propranolol, propafenone, oxyprenolol, nifedipine, reserpine, trimethaphan, phenoxybenzamine, pargyline hydrochloride, deserpidine, diazoxide, guanethidine monosulfate, minoxidil, rescinnamine, sodium nitroprusside, rauwolfia serpentina, alseroxylon, and phentolamine);
- 15 anti-inflammatories (e.g., (non-steroidal) indomethacin, ketoprofen, flurbiprofen, naproxen, ibuprofen, ramifenazone, piroxicam, (steroidal) cortisone, dexamethasone, fluazacort, celecoxib, rofecoxib, hydrocortisone, prednisolone, and prednisone);
- antineoplastics (e.g., cyclophosphamide, actinomycin, bleomycin, daunorubicin, doxorubicin, epirubicin, mitomycin, methotrexate, fluorouracil, carboplatin, carmustine
- 20 (BCNU), methyl-CCNU, cisplatin, etoposide, camptothecin and derivatives thereof, phenesterine, paclitaxel and derivatives thereof, docetaxel and derivatives thereof, vinblastine, vincristine, tamoxifen, and pipsulfan);
- antianxiety agents (e.g., lorazepam, buspirone, prazepam, chlordiazepoxide, oxazepam, clorazepate dipotassium, diazepam, hydroxyzine pamoate, hydroxyzine hydrochloride,
- 25 alprazolam, droperidol, halazepam, chlormezanone, and dantrolene);
- immunosuppressive agents (e.g., cyclosporine, azathioprine, mizoribine, and FK506 (tacrolimus));
- antimigraine agents (e.g., ergotamine, propranolol, isometheptene mucate, and dichloralphenazone);
- 30 sedatives/hypnotics (e.g., barbiturates such as pentobarbital, pentobarbital, and

- secobarbital; and benzodiazapines such as flurazepam hydrochloride, triazolam, and midazolam);
- antianginal agents (e.g., beta-adrenergic blockers; calcium channel blockers such as nifedipine, and diltiazem; and nitrates such as nitroglycerin, isosorbide dinitrate,
- 5 pentaerythritol tetranitrate, and erythrityl tetranitrate);
- antipsychotic agents (e.g., haloperidol, loxapine succinate, loxapine hydrochloride, thioridazine, thioridazine hydrochloride, thiothixene, fluphenazine, fluphenazine decanoate, fluphenazine enanthate, trifluoperazine, chlorpromazine, perphenazine, lithium citrate, and prochlorperazine);
- 10 antimanic agents (e.g., lithium carbonate);
- antiarrhythmics (e.g., bretylium tosylate, esmolol, verapamil, amiodarone, encainide, digoxin, digitoxin, mexiletine, disopyramide phosphate, procainamide, quinidine sulfate, quinidine gluconate, quinidine polygalacturonate, flecainide acetate, tocainide, and lidocaine);
- 15 antiarthritic agents (e.g., phenylbutazone, sulindac, penicillamine, salsalate, piroxicam, azathioprine, indomethacin, meclofenamate, gold sodium thiomalate, ketoprofen, aurano-fin, aurothioglucose, and tolmetin sodium);
- antigout agents (e.g., colchicine, and allopurinol);
- anticoagulants (e.g., heparin, heparin sodium, and warfarin sodium);
- 20 thrombolytic agents (e.g., urokinase, streptokinase, and alteplase);
- antifibrinolytic agents (e.g., aminocaproic acid);
- hemorheologic agents (e.g., pentoxifylline);
- antiplatelet agents (e.g., aspirin);
- anticonvulsants (e.g., valproic acid, divalproex sodium, phenytoin, phenytoin sodium,
- 25 clonazepam, primidone, phenobarbital, carbamazepine, amobarbital sodium, methsuximide, metharbital, mephobarbital, mephentyoin, phensuximide, paramethadione, ethotoin, phenacemide, secobarbital sodium, lorazepam dipotassium, and trimethadione);
- antiparkinson agents (e.g., ethosuximide);
- antihistamines/antipruritics (e.g., hydroxyzine, diphenhydramine, chlorpheniramine,
- 30 brompheniramine maleate, cyproheptadine hydrochloride, terfenadine, clemastine

- fumarate, triprolidine, carbinoxamine, diphenylpyraline, phenindamine, azatadine, tripeleminamine, dexchlorpheniramine maleate, and methdilazine);
- agents useful for calcium regulation (e.g., calcitonin, and parathyroid hormone);
- antibacterial agents (e.g., amikacin sulfate, aztreonam, chloramphenicol, chloramphenicol
- 5 palmitate, ciprofloxacin, clindamycin, clindamycin palmitate, clindamycin phosphate, metronidazole, metronidazole hydrochloride, gentamicin sulfate, lincomycin hydrochloride, tobramycin sulfate, vancomycin hydrochloride, polymyxin B sulfate, colistimethate sodium, and colistin sulfate);
- antiviral agents (e.g., interferon alpha, beta or gamma, zidovudine, amantadine
- 10 hydrochloride, ribavirin, and acyclovir);
- antimicrobials (e.g., cephalosporins such as cefazolin sodium, cephadrine, cefaclor, cephalixin sodium, ceftizoxime sodium, cefoperazone sodium, cefotetan disodium, cefuroxime azotil, cefotaxime sodium, cefadroxil monohydrate, cephalixin, cephalothin sodium, cephalixin hydrochloride monohydrate, cefamandole nafate, cefoxitin sodium,
- 15 cefonicid sodium, ceforanide, ceftriaxone sodium, ceftazidime, cefadroxil, cephradine, and cefuroxime sodium; penicillins such as ampicillin, amoxicillin, penicillin G benzathine, cyclacillin, ampicillin sodium, penicillin G potassium, penicillin V potassium, piperacillin sodium, oxacillin sodium, bacampicillin hydrochloride, cloxacillin sodium, ticarcillin disodium, azlocillin sodium, carbenicillin indanyl sodium, penicillin G procaine,
- 20 methicillin sodium, and nafcillin sodium; erythromycins such as erythromycin ethylsuccinate, erythromycin, erythromycin estolate, erythromycin lactobionate, erythromycin stearate, and erythromycin ethylsuccinate; and tetracyclines such as tetracycline hydrochloride, doxycycline hyclate, and minocycline hydrochloride, azithromycin, clarithromycin);
- 25 anti-infectives (e.g., GM-CSF);
- bronchodilators (e.g., sympathomimetics such as epinephrine hydrochloride, metaproterenol sulfate, terbutaline sulfate, isoetharine, isoetharine mesylate, isoetharine hydrochloride, albuterol sulfate, albuterol, bitolterolmesylate, isoproterenol hydrochloride, terbutaline sulfate, epinephrine, and epinephrine bitartrate, salbutamol, formoterol,
- 30 salmeterol, xinafoate, and pirbuterol);



steroidal compounds and hormones (e.g., androgens such as danazol, testosterone cypionate, fluoxymesterone, ethyltestosterone, testosterone enanthate, methyltestosterone, fluoxymesterone, and testosterone cypionate; estrogens such as estradiol, estropipate, and conjugated estrogens; progestins such as methoxyprogesterone acetate, and norethindrone acetate; corticosteroids such as triamcinolone, betamethasone, betamethasone sodium phosphate, dexamethasone, dexamethasone sodium phosphate, dexamethasone acetate, prednisone, methylprednisolone acetate suspension, triamcinolone acetonide, methylprednisolone, prednisolone sodium phosphate, methylprednisolone sodium succinate, hydrocortisone sodium succinate, triamcinolone hexacetonide, hydrocortisone, hydrocortisone cypionate, prednisolone, fludrocortisone acetate, paramethasone acetate, prednisolone tebutate, prednisolone acetate, prednisolone sodium phosphate, and hydrocortisone sodium succinate; and thyroid hormones such as levothyroxine sodium); hypoglycemic agents (e.g., human insulin, purified beef insulin, purified pork insulin, glyburide, chlorpropamide, glipizide, tolbutamide, and tolazamide); hypolipidemic agents (e.g., clofibrate, dextrothyroxine sodium, probucol, pravastatin, atorvastatin, lovastatin, and niacin); proteins (e.g., DNase, alginase, superoxide dismutase, and lipase); nucleic acids (e.g., sense or anti-sense nucleic acids encoding any therapeutically useful protein, including any of the proteins described herein); agents useful for erythropoiesis stimulation (e.g., erythropoietin); antiulcer/antireflux agents (e.g., famotidine, cimetidine, and ranitidine hydrochloride); antinauseants/antiemetics (e.g., meclizine hydrochloride, nabilone, prochlorperazine, dimenhydrinate, promethazine hydrochloride, thiethylperazine, ondasetron hydrochloride, palonsetron hydrochloride, and scopolamine); oil-soluble vitamins (e.g., vitamins A, D, E, K, and the like); as well as other pharmaceutical agents such as mitoxotranes, halonitrosoureas, anthrocyclines, and ellipticine. A description of these and other classes of useful pharmaceutical agents and a listing of species within each class can be found in Martindale, *The Extra Pharmacopoeia, 30th Ed.* (The Pharmaceutical Press, London 1993).

In one embodiment, the pharmaceutical agent comprises a corticosteroid.

Examples of corticosteroid include budesonide, fluticasone propionate, beclomethasone dipropionate, mometasone, flunisolide, and triamcinolone acetonide.

In another embodiment, the pharmaceutical agent comprises a bronchodilator. Examples of bronchodilators include albuterol, formoterol and salmeterol.

5 In another embodiment, the pharmaceutical agent comprises an antiasthmatic. Examples of antiasthmatics include, cromolyn sodium, and ipratropium bromide.

In a further embodiment, the pharmaceutical agent comprises another steroid, such as testosterone, progesterone, and estradiol.

In still another embodiment, the pharmaceutical agent comprises a leukotriene  
10 inhibitor (such as zafirlukast and zileuton), an antibiotic (such as cefprozil, ciprofloxacin, and amoxicillin), an antifungal (such as voriconazole and itraconazole), an antineoplastic (such as paclitaxel and docetaxel), or a peptide or protein (such as insulin, calcitonin, leuprolide, granulocyte colony-stimulating factor, parathyroid hormone-related peptide, growth hormone, interferons, erythropoietin, and somatostatin).

15 The content of pharmaceutical agent in the microparticles generally is between about 1 and about 70 wt%. In typical embodiments, the pharmaceutical agent is present in an amount between about 5 and 50 wt%.

In one embodiment, the sustained release formulations comprise two or more different pharmaceutical agents. In one embodiment, two or more pharmaceutical agents  
20 are combined into and delivered from one microparticle. In another embodiment, the formulation comprises a mixture of two or more different microparticles each containing a different pharmaceutical agent or pharmaceutical agents. In one embodiment, the formulation includes at least one pharmaceutical agent for sustained release and at least one other pharmaceutical agent for immediate release.

25 In yet another embodiment, the sustained release formulations comprise a mixture of different microparticles each containing a single pharmaceutical agent, but having different porosities, so that the some particles of the mixture have a first release profile (e.g., a majority of the first pharmaceutical agent is released between 2 and 6 hours) and other particles have a second pharmaceutical agent release profile (e.g., a majority of the  
30 second pharmaceutical agent is released between 6 and 12 hours, or between 6 and 24

hours).

### Materials To Inhibit Uptake by the RES

Uptake and removal of the microparticles by macrophages can be slowed or minimized through increasing the geometric particle size (e.g.,  $> 3\ \mu\text{m}$  slows phagocytosis) the selection of the polymer and/or incorporation or coupling of molecules that minimize adhesion or uptake or by incorporating the poly(alkylene glycol) into the matrix such that at least one glycol unit is surface exposed. For example, tissue adhesion by the microparticle can be minimized by covalently binding poly(alkylene glycol) moieties to the surface of the microparticle. The surface poly(alkylene glycol) moieties have a high affinity for water that reduces protein adsorption onto the surface of the particle. The recognition and uptake of the microparticle by the reticulo-endothelial system (RES) is therefore reduced.

In one method, the terminal hydroxyl group of the poly(alkylene glycol) is covalently attached to biologically active molecules, or molecules affecting the charge, lipophilicity or hydrophilicity of the particle, onto the surface of the microparticle. Methods available in the art can be used to attach any of a wide range of ligands to the microparticles to enhance the delivery properties, the stability or other properties of the microparticles *in vivo*.

### Bulking Agents

For administration to the pulmonary system using a dry powder inhaler, the porous microparticles can be combined (e.g., blended) with one or more pharmaceutically acceptable bulking agents and administered as a dry powder. Examples of pharmaceutically acceptable bulking agents include sugars such as mannitol, sucrose, lactose, fructose and trehalose and amino acids. Amino acids that can be used include glycine, arginine, histidine, threonine, asparagine, aspartic acid, serine, glutamate, proline, cysteine, methionine, valine, leucine, isoleucine, tryptophan, phenylalanine, tyrosine, lysine, alanine, and glutamine. In one embodiment, the bulking agent comprises particles having a volume average size between 10 and 500  $\mu\text{m}$ .

### Suspending Agents

For administration to the pulmonary system, the porous microparticles can be suspended with one or more pharmaceutically acceptable suspending agents that are liquid

within a metered dose inhaler and administered via a metered dose inhaler. Examples of pharmaceutically acceptable suspending agents include chlorofluorocarbons and hydrofluorocarbons. Examples of pharmaceutically acceptable suspending agent for use in metered dose inhalers include hydrofluorocarbons (such as HFA-134a and HFA-227) and chlorofluorocarbons (such as CFC-11, CFC-12, and CFC-114). Mixtures of suspending agents can be used.

#### **Making the Porous Microparticles and Sustained Release Formulations**

In typical embodiments, the porous microparticles are made by a method that includes the following steps: (1) dissolving the matrix material in a volatile solvent to form a matrix material solution; (2) adding the pharmaceutical agent to the solution of matrix material; (3) optionally combining at least one pore forming agent with the pharmaceutical agent in the matrix material solution and emulsifying to form an emulsion, suspension, or second solution; and (4) removing the volatile solvent, and the pore forming agent if present, from the emulsion, suspension, or second solution to yield porous microparticles which comprise the pharmaceutical agent and the matrix material. The method produces microparticles that upon inhalation of the formulation into the lungs release a therapeutically or prophylactically effective amount of the pharmaceutical agent from the microparticles in the lungs for at least 2 hours. Techniques that can be used to make the porous microparticles include melt extrusion, spray drying, fluid bed drying, solvent extraction, hot melt encapsulation, and solvent evaporation, as discussed below. In the most preferred embodiment, microparticles are produced by spray drying. The pharmaceutical agent can be incorporated into the matrix as solid particles, liquid droplets, or by dissolving the pharmaceutical agent in the matrix material solvent. If the pharmaceutical agent is a solid, the pharmaceutical agent may be encapsulated as solid particles which are added to the matrix material solution or may be dissolved in an aqueous solution which then is emulsified with the matrix material solution prior to encapsulation, or the solid pharmaceutical agent may be cosolubilized together with the matrix material in the matrix material solvent.

In one embodiment, the method further comprises combining one or more surfactants, with the pharmaceutical agent in a matrix material solution. In one

embodiment of the methods for making sustained release formulations, the process further includes blending the porous microparticles with a pharmaceutically acceptable bulking agent.

In one example, the matrix material comprises a biocompatible synthetic polymer, and the volatile solvent comprises an organic solvent. In another example, the pore forming agent is in the form of an aqueous solution when combined with the pharmaceutical agent/matrix solution.

In one embodiment, the step of removing the volatile solvent and pore forming agent from the emulsion, suspension, or second solution is conducted using a process selected from spray drying, evaporation, fluid bed drying, lyophilization, vacuum drying, or a combination thereof.

#### Solvent Evaporation

In this method, the matrix material and pharmaceutical agent are dissolved in a volatile organic solvent such as methylene chloride. A pore forming agent as a solid or as a liquid may be added to the solution. The active agent can be added as either a solid or in solution to the polymer solution. The mixture is sonicated or homogenized and the resulting dispersion or emulsion is added to an aqueous solution that may contain a surface active agent such as TWEEN™ 20, TWEEN™ 80, PEG or poly(vinyl alcohol) and homogenized to form an emulsion. The resulting emulsion is stirred until most of the organic solvent evaporates, leaving microparticles. Microparticles with different geometric sizes and morphologies can be obtained by this method by controlling the emulsion droplet size. Solvent evaporation is described by Mathiowitz, et al., J. Scanning Microscopy, 4:329 (1990); Beck, et al., Fertil. Steril., 31:545 (1979); and Benita, et al., J. Pharm. Sci., 73:1721 (1984).

Particularly hydrolytically unstable polymers, such as polyanhydrides, may degrade during the fabrication process due to the presence of water. For these polymers, the following two methods, which are performed in completely organic solvents, are more useful.

#### Hot Melt Microencapsulation

In this method, the matrix material and the pharmaceutical agent are first melted

and then mixed with the solid or liquid active agent. A pore forming agent as a solid or in solution may be added to the solution. The mixture is suspended in a non-miscible solvent (like silicon oil), and, while stirring continuously, heated to 5 °C above the melting point of the polymer. Once the emulsion is stabilized, it is cooled until the polymer particles solidify. The resulting microparticles are washed by decantation with a polymer non-solvent such as petroleum ether to give a free-flowing powder. Hot-melt microencapsulation is described by Mathiowitz, et al., Reactive Polymers, 6:275 (1987).

#### Solvent Removal

This technique was primarily designed for hydrolytically unstable materials. In this method, the solid or liquid pharmaceutical agent is dispersed or dissolved in a solution of the selected matrix material and pharmaceutical agent in a volatile organic solvent like methylene chloride. This mixture is suspended by stirring in an organic oil (such as silicon oil) to form an emulsion. The external morphology of particles produced with this technique is highly dependent on the type of polymer used.

#### Spray Drying of Microparticles

Microparticles can be produced by spray drying by a method that includes the following steps: (1) dissolving the matrix material, and optionally a surfactant, in a volatile solvent to form a matrix material solution; (2) adding a pharmaceutical agent to the solution of matrix material; (3) optionally combining at least one pore forming agent with the pharmaceutical agent in the matrix material solution; (4) forming an emulsion, suspension or second solution from the pharmaceutical agent, the matrix material solution, and the optional pore forming agent; and (5) spray drying the emulsion, suspension or solution and removing the volatile solvent and the pore forming agent, if present, to form porous microparticles. As defined herein, the process of "spray drying" an emulsion, suspension or solution containing a matrix material and a pharmaceutical agent refers to a process wherein the emulsion, suspension or solution is atomized to form a fine mist and dried by direct contact with temperature-controlled carrier gases. In a typical embodiment using spray drying apparatus available in the art, the emulsion, suspension or solution is delivered through the inlet port of the spray drier, passed through a tube within the drier and then atomized through the outlet port. The temperature may be varied depending on

the gas or matrix material used. The temperature of the inlet and outlet ports can be controlled to produce the desired products.

The geometric size of the particulates formed is a function of the atomizer used to spray the matrix material solution, atomizer pressure, the flow rate, the matrix material used, the matrix material concentration, the type of solvent and the temperature of spraying (both inlet and outlet temperature). Microparticles ranging in geometric diameter between one and ten microns can be obtained.

If the pharmaceutical agent is a solid, the agent may be encapsulated as solid particles which are added to the matrix material solution prior to spraying, or the pharmaceutical agent can be dissolved in a solvent which then is emulsified with the matrix material solution prior to spraying, or the solid may be cosolubilized together with the matrix material in an appropriate solvent prior to spraying.

#### Reagents for Making the Porous Microparticles

Certain reagents used to make the porous microparticles may include solvents for the matrix material, solvents or vehicles for the pharmaceutical agent, pore forming agents, and various additives to facilitate microparticle formation.

#### Solvents

A solvent for the matrix material is selected based on its biocompatibility as well as the solubility of the matrix material and where appropriate, interaction with the pharmaceutical agent to be delivered. For example, the ease with which the matrix material is dissolved in the solvent and the lack of detrimental effects of the solvent on the pharmaceutical agent to be delivered are factors to consider in selecting the matrix material solvent. Aqueous solvents can be used to make matrices formed of water-soluble polymers. Organic solvents will typically be used to dissolve hydrophobic and some hydrophilic matrix materials. Combinations of aqueous and organic solvents may be used. Preferred organic solvents are volatile or have a relatively low boiling point or can be removed under vacuum and which are acceptable for administration to humans in trace amounts, such as methylene chloride. Other solvents, such as ethyl acetate, ethanol, methanol, dimethyl formamide (DMF), acetone, acetonitrile, tetrahydrofuran (THF), acetic acid, dimethyl sulfoxide (DMSO) and chloroform, and combinations thereof, also may be

utilized. Preferred solvents are those rated as class 3 residual solvents by the Food and Drug Administration, as published in the Federal Register vol. 62, number 85, pp. 24301-09 (May 1997).

5 In general, the matrix material is dissolved in the solvent to form a matrix material solution having a concentration of between 0.1 and 60% weight to volume (w/v), more preferably between 0.25 and 30%. The matrix material solution is then processed as described below to yield a matrix having pharmaceutical agents incorporated therein.

#### Surfactants to Facilitate Microparticle Formation

10 A variety of surfactants may be added to a solution, suspension, or emulsion containing matrix material to facilitate microparticle formation. The surfactants may be added to any phase of an emulsion as emulsifiers if an emulsion is used during the production of the matrices. Exemplary emulsifiers or surfactants that may be used (e.g., between about 0.1 and 5 % by weight relative to weight of the pharmaceutical agent and matrix material) include most physiologically acceptable emulsifiers. Examples include  
15 natural and synthetic forms of bile salts or bile acids, both conjugated with amino acids and unconjugated such as taurodeoxycholate, and cholic acid. Phospholipids can be used as mixtures, including natural mixtures such as lecithins. These surfactants may function solely as emulsifiers, and as such form part of and are dispersed throughout the matrix of the particles.

#### 20 Additives to Facilitate Microparticle Dispersion

The composition of the microparticles may comprise a surfactant in a manner such that the microparticles will have all or part of the surfactant structure surface exposed, and as such will facilitate dispersion of the microparticles for administration via dry powder inhaler or via metered dose inhaler. Surfactants for facilitating dispersion may be included  
25 during production of the microparticles. Alternatively, the microparticles may be coated with the surfactant post-production. Exemplary surfactants that may be used (e.g., between about 0.1 and 5 % by weight relative to weight of the pharmaceutical agent and matrix material) include phospholipids, salts of fatty acids, and molecules containing PEG units such as polysorbate 80.



### Control of Porosity

The porosity of the microparticles can be controlled during the production of the microparticles by adjusting the solids content of the pharmaceutical agent in matrix material solution or adjusting the rate at which the matrix solvent is removed, or  
5 combinations thereof. Higher solids concentrations lead to microparticles with less porosity.

Alternatively, pore forming agents as described below can be used to control the porosity of the microparticles during production. Pore forming agents are volatile materials that are used during the process to create porosity in the resultant matrix. The  
10 pore forming agent can be a volatilizable solid or volatilizable liquid.

#### Liquid Pore Forming Agent

The liquid pore forming agent must be immiscible with the matrix material solvent and volatilizable under processing conditions compatible with the pharmaceutical agent and matrix material. To effect pore formation, the pore forming agent first is emulsified  
15 with the pharmaceutical agent in the matrix material solution. Then, the emulsion is further processed to remove the matrix material solvent and the pore forming agent simultaneously or sequentially using evaporation, vacuum drying, spray drying, fluid bed drying, lyophilization, or a combination of these techniques.

The selection of liquid pore forming agents will depend on the matrix material  
20 solvent. Representative liquid pore forming agents include water; dichloromethane; alcohols such as ethanol, methanol, or isopropanol; acetone; ethyl acetate; ethyl formate; dimethylsulfoxide; acetonitrile; toluene; xylene; dimethylformamide; ethers such as THF, diethyl ether, or dioxane; triethylamine; formamide; acetic acid; methyl ethyl ketone; pyridine; hexane; pentane; furan; water; liquid perfluorocarbons, and cyclohexane.

25 The liquid pore forming agent is used in an amount that is between 1 and 50% (v/v), preferably between 5 and 25% (v/v), of the pharmaceutical agent solvent emulsion.

#### Solid Pore Forming Agent

The solid pore forming agent must be volatilizable under processing conditions which do not harm the pharmaceutical agent or matrix material. The solid pore forming  
30 agent can be (i) dissolved in the matrix material solution which contains the

pharmaceutical agent, (ii) dissolved in a solvent which is not miscible with the matrix material solvent to form a solution which is then emulsified with the matrix material solution which contains the pharmaceutical agent, or (iii) added as solid particulates to the matrix material solution which contains the pharmaceutical agent. The solution, emulsion, or suspension of the pore forming agent in the pharmaceutical agent/matrix material solution then is further processed to remove the matrix material solvent, the pore forming agent, and, if appropriate, the solvent for the pore forming agent simultaneously or sequentially using evaporation, spray drying, fluid bed drying, lyophilization, vacuum drying, or a combination of these techniques. After the matrix material is precipitated, the hardened microparticles can be frozen and lyophilized to remove any pore forming agents not removed during the microencapsulation process.

In a preferred embodiment, the solid pore forming agent is a volatile salt, such as salts of volatile bases combined with volatile acids. Volatile salts are materials that can transform from a solid or liquid to a gaseous state using added heat and/or vacuum. Examples of volatile bases include ammonia, methylamine, ethylamine, dimethylamine, diethylamine, methylethylamine, trimethylamine, triethylamine, and pyridine. Examples of volatile acids include carbonic acid, hydrochloric acid, hydrobromic acid, hydroiodic acid, formic acid, acetic acid, propionic acid, butyric acid, and benzoic acid. Preferred volatile salts include ammonium bicarbonate, ammonium acetate, ammonium chloride, ammonium benzoate and mixtures thereof. Other examples of solid pore forming agents include iodine, phenol, benzoic acid (as acid not as salt), camphor, and naphthalene.

The solid pore forming agent is used in an amount between 5 and 1000% (w/w), preferably between 10 and 600% (w/w), and more preferably between 10 and 100% (w/w), of the pharmaceutical agent and the matrix material.

## **Methods of Administering the Porous Microparticles**

The sustained release formulation comprising porous microparticles as described herein preferably is administered to the lungs of a patient by oral inhalation, for example by having the patient inhale a dry powder form of the formulation using a suitable inhalation device. Dry powder inhalation devices for medicaments, which disperse the pharmaceutical agent in air or a propellant, are well known in the art. See, e.g., U.S. Patent

No. 5,327,883; No. 5,577,497; and No. 6,060,069. Types of inhalation devices include dry powder inhalers (DPIs), metered dose inhalers (MDIs), and nebulizers. Commercial embodiments of some of these include the SPIROS<sup>TM</sup> DPI (Dura Pharmaceuticals, Inc. US), the ROTOHALER<sup>TM</sup>, the TURBUHALER<sup>TM</sup> (Astra SE), the CYCLOHALER<sup>TM</sup> (Pharmachemie B.V.), FLOWCAPS<sup>TM</sup> (Hovione) and the VENTODISK<sup>TM</sup> (Glaxo, UK).  
5 For administration to the pulmonary system using a dry powder inhaler, the porous microparticles can be combined (e.g., blended) with one or more pharmaceutically acceptable bulking agents and administered as a dry powder. Examples of pharmaceutically acceptable bulking agents include sugars such as mannitol, sucrose,  
10 lactose, fructose, and trehalose and amino acids.

In one embodiment, the sustained release formulation with or without bulking agent is loaded into a unit dose receptacle (e.g., a gelatin, hydropropylmethylcellulose or plastic capsule, or blister) which is then placed within a suitable inhalation device to allow for the aerosolization of the dry powder formulation by dispersion into a gas stream to form an  
15 aerosol, which is captured in a chamber having an attached mouthpiece. The patient can inhale the aerosol through the mouthpiece to initiate pharmaceutical agent delivery and treatment.

In another embodiment, the sustained release formulation comprises one or more pharmaceutically acceptable suspending agents that are liquid within a conventional  
20 metered dose inhaler to form a metered dose inhaler formulation. Examples of pharmaceutically acceptable suspending agents for use in metered dose inhalers are hydrofluorocarbons (such as HFA-134a, and HFA-227) and chlorofluorocarbons (such as CFC-11, CFC-12 and CFC-114). Mixtures of the suspending agents may be used.

### **Treatments**

25 The sustained release formulations are useful in a variety of inhalation pharmaceutical agent delivery applications. The applications can be for local delivery and treatment of the lungs, or for systemic delivery via the lungs (for any treatment or prophylaxis). Relative to systemic pharmaceutical agent delivery via the oral or injectable route, local delivery of respiratory pharmaceutical agents via the pulmonary route requires  
30 smaller doses of the pharmaceutical agent and minimizes systemic toxicity because it can

be delivered directly to the site of the disease.

In one embodiment, the sustained release formulations are useful in the treatment of a respiratory disease. Examples include asthma, COPD, cystic fibrosis, and lung cancer.

In one embodiment, administration of the sustained release formulations described  
5 herein provides local or plasma concentrations sustained at approximately constant values over the intended period of release (e.g., up to 2 to 24 hours, to enable twice- to once-daily dosing). The sustained release formulations may allow patients to take treatments for such diseases as asthma less frequently, and to receive more prolonged and steadier relief.

The methods and compositions described above will be further understood with  
10 reference to the following non-limiting examples.

### **Examples**

In the examples below, where porosity of microparticles was determined, the following procedure was used: TAP Density (Transaxial Pressure Density as a measure of tap density) for the microparticles was determined using a Micromeritics GeoPyc Model  
15 1360. Envelope density for the microparticles was estimated from the TAP density (EQ.5). Absolute density was determined via helium pycnometry using a Micromeritics AccuPyc Model 1330. The absolute densities of the polymer, pharmaceutical agent, and phospholipid were determined, and a weighted average value was used for the absolute density of the microparticles. The porosity was calculated based on EQ.6 above. Where  
20 percent porosity is reported, the value of porosity (based on EQ.6) was multiplied by 100%.

In the examples below, the *in vitro* pharmaceutical agent release rate was determined using the following procedure. Microparticles were suspended in PBS-SDS (Phosphate Buffered Saline - 0.05% Sodium Dodecyl Sulfate) such that the nominal  
25 pharmaceutical agent concentration in the suspension was 1 mg/mL. A sample of the suspension was then added to a large volume of PBS-SDS at 37 °C, such that theoretical pharmaceutical agent concentration at 100% release was 0.75 µg/mL. The resulting diluted suspension was maintained at 37 °C in an incubator on a rocker. To determine the release rate of pharmaceutical agent from the microparticles, samples of the release media were  
30 taken over time, the microparticles separated from the solution, and the solution

pharmaceutical agent concentration was monitored via HPLC with detection at 254 nm for budesonide or 238 nm for fluticasone propionate. The column was a J'Sphere ODS-H80 (250 x 4.6 mm, 4 µm). The mobile phase was an isocratic system consisting of Ethanol-Water (64:36), running at a flow rate of 0.8 mL/min.

5            In the examples below, where geometric particle size is described, the volume average size was measured using a Coulter Multisizer II with a 50 µm aperture.

Powders were dispersed in an aqueous vehicle containing Pluronic F127 and mannitol using vortexing and sonication. The resulting suspensions were then diluted into electrolyte for analysis.

10

#### **Example 1:    Effect of Microparticle Porosity on Budesonide Release**

Microspheres containing budesonide were prepared, using materials obtained as follows: budesonide was from FarmaBios S.R.L. (Pavia, Italy); phospholipid (DPPC) was from Avanti Polar Lipids Inc. (Alabaster, AL); polymer (PLGA) was from BI Chemicals  
15 (Petersburg, VA); ammonium bicarbonate was from Spectrum Chemicals (Gardena, CA); and methylene chloride was from EM Science (Gibbstown, NJ).

Six different lots of budesonide containing microspheres (B1 through B6) were prepared as follows. For each microsphere lot (B1-B4 and B6) 8.0 g of PLGA, 0.72 g of DPPC, and 2.2 g of budesonide were dissolved into 364 mL of methylene chloride at 20  
20 °C. For lot B5, 36.0 g of PLGA, 2.16 g of DPPC, and 9.9 g of budesonide were dissolved into 1764 mL of methylene chloride at 20 °C. Lot B1 was prepared without a pore forming agent, and the process conditions and solids content of the solution to the spray dryer were used to create the porosity of the microspheres. Lots B2-B6 were prepared using the pore forming agent, ammonium bicarbonate to create microspheres having porosities greater  
25 than lot B1. For lots B2-B6, a stock solution of the pore forming agent was prepared by dissolving 4.0 g of ammonium bicarbonate into 36 mL of RO/DI water at 20 °C. For each lot, a different ratio of the ammonium bicarbonate stock solution was combined with the pharmaceutical agent/polymer solution (volume pore forming agent: pharmaceutical agent/polymer solution: B2: 1:49, B3: 1:24, B4: 1:10, B5: 1:49, B6: 1:19) described above  
30 and emulsified using a rotor-stator homogenizer. The resulting emulsion was spray dried on a benchtop spray dryer using an air-atomizing nozzle and nitrogen as the drying gas.

Spray drying conditions were as follows: 20 mL/min emulsion flow rate, 60 kg/hr drying gas rate and 21 °C outlet temperature. The product collection container was detached from the spray dryer and attached to a vacuum pump, where it was dried for at least 18 hours.

FIG.1 is a graph of percent of budesonide released *in vitro* after 5.5 hours versus porosity. Table 1 shows the geometric size, density and porosity data for the lots shown in FIG. 1.

**Table 1: Geometric Size, Tap Density and Porosity  
Of the Budesonide-Containing Microspheres**

Lot #	Geometric Size ( $\mu\text{m}$ )	Tap density (g/mL)	Porosity x 100 %
B4	2.3	0.22	81
B3	2.1	0.44	61
B2	2.5	0.53	53
B1	1.7	0.68	40

Table 2 further illustrates the effect of porosity on the percent budesonide released after 24 hours.

**Table 2: Effect of Porosity on Budesonide Release After 24 Hours**

Lot #	Porosity x 100 %	% Budesonide release after 24 hours
B6	57.8	86.5
B5	46.1	58.9

The *in vitro* budesonide release data demonstrate how the control of porosity can be used to adjust the amount of pharmaceutical agent released after a certain period of time, and how porosity can be used to ensure that significant release of the pharmaceutical agent occurs beyond the initial release and that the majority of the pharmaceutical agent is released within 24 hours.

#### **Example 2: Effect of Microparticle Porosity on Fluticasone Propionate Release**

Microspheres containing fluticasone propionate were prepared, using materials obtained as follows: fluticasone propionate was from Cipla Ltd. (Mumbai, India); phospholipid (DPPC) was from Chemi S.p.A. (Milan, Italy); polymer (PLGA) was from BI

Chemicals (Petersburg, VA); ammonium bicarbonate was from Spectrum Chemicals (Gardena, CA); and methylene chloride was from EM Science (Gibbstown, NJ).

Six different lots of fluticasone propionate containing microspheres (F1 through F6) were prepared as follows. For each microsphere lot, 3.0 g of PLGA, 0.18 g of DPPC, and 0.825 g of fluticasone propionate were dissolved into 136.4 mL of methylene chloride at 20 °C. Lot F1 was prepared without a pore forming agent, and the process conditions and solids content of the solution to the spray dryer were used to create the porosity of the microspheres. Lots F2-F6 were prepared using the pore forming agent ammonium bicarbonate to create microspheres having porosities greater than lot F1. A stock solution of the pore forming agent was prepared by dissolving 2.22 g of ammonium bicarbonate into 20 g of RO/DI water at 20 °C. For each lot, a different ratio of ammonium bicarbonate stock solution was combined with the pharmaceutical agent/polymer solution (volume ammonium bicarbonate solution: volume pharmaceutical agent/polymer solution: F2: 1:74, F3: 1:49, F4: 1:24, F5: 1:14, F6: 1:10) and the mixture was then emulsified using a rotor-stator homogenizer. The resulting emulsion was spray dried on a benchtop spray dryer using an air-atomizing nozzle and nitrogen as the drying gas. Spray drying conditions were as follows: 20 mL/min emulsion flow rate, 60 kg/hr drying gas rate, and 21 °C outlet temperature. The product collection container was detached from the spray dryer and attached to a vacuum pump, where it was dried for at least 18 hours.

FIGS. 2 and 3 are graphs of percent of fluticasone released *in vitro* after 5.5 hours and 24 hours, respectively, versus porosity. Table 3 shows the geometric size, density, and porosity data for the material whose release is shown in FIGS. 2 and 3.

**Table 3: Geometric Size, Tap Density, and Porosity  
Of the Fluticasone Propionate-Containing Microspheres**

Lot #	Geometric Size (μm)	Tap density (g/mL)	Porosity x 100 %
F6	3.8	0.31	73
F5	3.5	0.31	73
F4	3.4	0.56	51
F3	2.7	0.59	48
F2	3.1	0.72	37
F1	3.1	0.82	28

The *in vitro* fluticasone propionate release data demonstrate how porosity can be used to

adjust the amount of pharmaceutical agent released after a certain period of time and can be used to ensure that significant release of the pharmaceutical agent.

**Example 3: Production of Radiolabeled Budesonide-Containing Microspheres  
For a Human Clinical Study**

5 Budesonide-containing microspheres were produced in manner similar to lot B5 described in Example 1, using materials obtained as follows: budesonide was from FarmaBios S.R.L. (Pavia, Italy); phospholipid (DPPC) was from Chemi S.p.A. (Milan, Italy); polymer (PLGA) was from BI Chemicals (Petersburg, VA); ammonium bicarbonate  
10 was from Spectrum Chemicals (Gardena, CA); methylene chloride was from EM Science (Gibbstown, NJ), lactose (325M) was from DMV (Veghel, The Netherlands), and gelatin capsules (size 3, Coni-Snap) were from Capsugel (Greenwood, SC).

A solution was prepared by dissolving 8.0 g of PLGA, 2.2 g of budesonide, and 0.48 g of DPPC in 392 mL of methylene chloride at 20 °C. A solution of the pore forming  
15 agent was prepared by dissolving 1.11 g of ammonium bicarbonate in 10 mL of distilled water at 20 °C. Eight milliliter of the aqueous solution was added to the organic solution and homogenized. The resulting emulsion was spray dried on a benchtop spray dryer using an air-atomizing nozzle and nitrogen as the drying gas. Spray drying conditions were as follows: 20 ml/min solution flow rate, 60 kg/hr drying gas rate, and 21 °C outlet  
20 temperature. The product collection container was detached from the spray dryer and attached to a vacuum pump, where it was dried for at least 24 hours.

The dried microspheres were then radiolabeled with technetium. The radiolabeled microspheres were transferred to a stainless steel mixing vessel and manually mixed with lactose. The mixed materials were then blended on a Turbula shaker-mixer, and the  
25 blended material was manually filled into gelatin capsules, giving a nominal pharmaceutical agent loading of 824 µg/capsule.

**Example 4: Administration of Budesonide-Containing Microspheres  
To Human Subjects by Inhalation**

30 A randomized, open-label, single-dose, single-centre, crossover study in healthy volunteers (10 subjects) was conducted comparing pharmacokinetics and pulmonary



deposition of the budesonide microspheres produced in Example 3 delivered by dry powder inhaler (Rotahaler, Glaxo Smith Kline, 3 actuations per subject) and an immediate release budesonide formulation delivered using a commercial dry powder inhaler (Pulmicort Turbuhaler, 4 actuations per subject, 200 µg/actuation). The doses administered for both formulations were significantly higher than would be administered under therapeutic conditions, to ensure plasma levels of budesonide above the level of detection and thus allow the *in vivo* release profile of the microspheres to be assessed. Plasma concentrations of budesonide were measured at 0, 2, 4, 6, 8, 12, 20, 30, 45, 60 minutes, and 1.5, 2, 3, 4, 6, 8, 10 and 12 hours after the final inhalation of each dosing period. Plasma samples were analyzed using a validated LC/MS/MS method. The plasma profiles adjusted for actual inhaled dose are shown in FIG. 4. The average values for the 10 subjects are reported.

Non-compartmental analysis was performed on the plasma curves. The results indicated a significant difference in the budesonide mean absorption time following inhalation ( $MAT_{inh}$ ) of 2.5 hrs for the immediate release formulation (Pulmicort) as compared to 10 hrs for the budesonide-containing microsphere preparation, as shown in Table 4 (the average and the standard deviation for the 10 subjects are reported). This clearly indicates that budesonide was absorbed slowly into the systemic circulation after inhalation of the budesonide microsphere as compared to inhalation of the immediate release formulation. The microspheres provided a four-fold increase in MAT as compared to the immediate release Pulmicort budesonide formulation.

**Table 4: MAT Comparisons of the Budesonide Formulations Following Inhalation**

Pharmaceutical Agent	$MAT_{inh}$ (hours)
Pulmicort (commercial product)	$2.5 \pm 1.8$
Budesonide-containing Microsphere Formulation (produced in Example 3)	$10.1 \pm 4.1$

The regional distribution of the microspheres in the lung was determined via gamma scintigraphy. Approximately 80% of inhaled microspheres (made and blended in Example 3) was delivered to the intended target, the upper lung. The microspheres remained in the lung for up to 24 hours, the period of time required for once-daily dosing.

Publications cited herein and the materials for which they are cited are specifically incorporated by reference. Modifications and variations of the methods and devices described herein will be obvious to those skilled in the art from the foregoing detailed description. Such modifications and variations are intended to come within the scope of  
5 the appended claims.